

What is claimed is:

1. A method for the production of para-hydroxy benzoic acid in a green plant comprising:

- 5           a) providing a green plant having an endogenous source of chorismate and containing a chorismate pyruvate lyase expression cassette having the following structure:

P-T-C-D-CPL

wherein:

10           P is a promoter suitable for driving the expression of a chorismate pyruvate lyase gene;

15           T is a nucleic acid molecule encoding a rubisco chloroplast transit peptide;

20           C a nucleic acid molecule encoding a Rubisco chloroplast transit peptide cleavage site;

25           D is a nucleic acid molecule encoding from about 4 to about 20 contiguous amino acids of the N-terminal portion of a Rubisco chloroplast transit peptide donor polypeptide; and

30           CPL is a nucleic acid molecule encoding a mature chorismate pyruvate lyase protein;

35           wherein each of P, T, C, D, and CPL are operably linked such that expression of the cassette results in translation of a chimeric protein comprising a chloroplast targeting sequence fused to the N-terminus of the mature chorismate pyruvate lyase protein;

          b) growing said plant under conditions whereby the chimeric protein is expressed and translocated to the chloroplast for the conversion of chorismate to para-hydroxy benzoic acid glucoside and para-hydroxy benzoic acid derivatives;

          c) recovering para-hydroxy benzoic acid and para-hydroxy benzoic acid derivatives from the plant; and

          d) processing said the para-hydroxy benzoic acid glucoside and para-hydroxy benzoic acid derivatives to free para-hydroxy benzoic acid.

2. A method according to Claim 1 wherein the Rubisco transit peptide is derived from a plant selected from the group consisting of: soybean, rapeseed, sunflower, cotton, corn, tobacco, alfalfa, wheat, barley, oats, sorghum, rice, *Arabidopsis*, sugar beet, sugar cane, canola, millet, beans, peas, rye, flax, and forage grasses.

3. A method according to Claim 1 wherein the promoter is selected from the group consisting of the 35S promoter, the nopaline synthase promoter, the octopine synthase promoter, cauliflower mosaic virus promoter, the ribulose-1,5-bisphosphate carboxylase promoter and the promoter of the chlorophyll a/b binding protein.
- 5 4. A method according to Claim 1 wherein the chorismate pyruvate lyase enzyme is encoded by the nucleic acid sequence as set forth in SEQ ID NO:3.
5. A method according to Claim 1 wherein the chimeric protein comprising a chloroplast targeting sequence fused to the N-terminus of the mature chorismate pyruvate lyase protein has the amino acid sequence as set forth in SEQ ID NO:8.
- 10 6. A method according to Claim 5 wherein the chimeric protein comprising a chloroplast targeting sequence fused to the N-terminus of the mature chorismate pyruvate lyase protein is processed to the amino acid sequence as set forth in SEQ ID NO:16.
- 15 7. A method according to Claim 1 wherein the pHBA glucoside is produced at a concentration of a least 2% para-hydroxy benzoic acid glucoside per dry weight of plant biomass.
8. A method according to Claim 1 wherein the para-hydroxy benzoic acid glucoside is produced at a concentration of at least 10% para-hydroxy benzoic acid glucoside per dry weight of plant biomass.
- 20 9. A method according to Claim 1 wherein the green plant containing a chorismate pyruvate lyase expression cassette is selected from the group consisting of soybean, rapeseed, sunflower, cotton, corn, tobacco, alfalfa, wheat, barley, oats, sorghum, rice, *Arabidopsis*, sugar beet, sugar cane, canola, millet, beans, peas, rye, flax, and forage grasses.
- 25 10. A method according to Claim 1 wherein the para-hydroxy benzoic acid is produced at a concentration of greater than 4.5% pHBA per dry weight of plant biomass.
- 30 11. A chorismate pyruvate lyase expression cassette comprising a chimeric gene having a nucleic acid molecule encoding a ribulose-1,5-bisphosphate carboxylase small subunit chloroplast targeting sequence having an amino acid sequence as set forth in SEQ ID NO:15 operably linked to a nucleic acid molecule encoding a chorismate pyruvate lyase enzyme having the amino acid sequence as set forth in SEQ ID NO:4.
- 35 12. A chorismate pyruvate lyase expression cassette according to Claim 11 wherein the chimeric gene encodes a polypeptide as set forth in SEQ ID NO:8.

13. A plant comprising the CPL expression cassette of Claim 11.
14. The plant according to Claim 13 selected from the group consisting of soybean, rapeseed, sunflower, cotton, corn, tobacco, alfalfa, wheat, barley, oats, sorghum, rice, *Arabidopsis*, sugar beet, sugar cane, canola, millet, beans, peas, rye, flax, and forage grasses.
- 5      15. A chimeric protein comprising a chloroplast targeting sequence fused to the N-terminus of the mature chorismate pyruvate lyase protein having the amino acid sequence as set forth in SEQ ID NO:8.
16. An isolated nucleic acid fragment encoding a chimeric protein
- 10     comprising a chloroplast targeting sequence fused to the N-terminus of the mature CPL protein has the amino acid sequence as set forth in SEQ ID NO:15.
17. An isolated nucleic acid fragment of Claim 16 having the sequence as set forth in SEQ ID NO:7.
18. The chloroplast cleavage product of Claim 15 having the amino acid sequence as set forth in SEQ ID NO:16.
- 15     19. A nucleic acid fragment encoding the processed protein of Claim 18.